L8: Entry 5 of 33 File: USPT Oct 1, 2002

DOCUMENT-IDENTIFIER: US 6458599 B1
TITLE: Compositions and methods for capturing, isolating, detecting, analyzing and quantifying macromolecules

Detailed Description Text (64):
Once the matrix is in a solid or semi-solid state, the molecular imprints can be processed to take on a variety of shapes. Usually, the molecular imprint will initially take on the same shape as the container used to create matrix 14'. However, any shape that might be useful for capturing macromolecules is possible. For example, they may be in the form of individual beads, disks, ellipses, or other regular or irregular shapes (collectively referred to as "beads"), or in the form of sheets. Beads can be formed by grinding a rigid matrix 14' or by suspension and dispersion techniques. Methods of making imprinted beads are discussed in Damen et al., 1980, J. Am. Chem. Soc. 102:3265-3267; Braun et al., 1984, Chemiker-Zeitung 108:255-257; and Bystrom et al., 1993, J. Am. Chem. Soc. 115:2081-2083. Imprinted beads may also be prepared by imprinting in the pore network of preformed beaded silica as discussed in Wulff et al., 1985, Reactive Polymers 3:261-2757. Dispersion techniques are discussed in Sellergren et al., 1994, 673:133-141. The formation of beaded molecular imprints by suspension polymerization is described in U.S. Patent No. 5,821,311. All of these references are incorporated herein by reference.

Detailed Description Text (116):
Acrylamide monomer solution was prepared by dissolving 28.5 g acrylamide and 1.5 g N-N'-methylene bisacrylamide in 100 ml of 4 M urea. 2 mg of the palmitoyl-peptide conjugate molecule of Example 1 was dissolved in 1 ml of the acrylamide monomer solution. Ammonium persulfate and TEMED were added to the solution as catalysts. The final concentration of ammonium persulfate was 0.02%, and the final concentration of TEMED was 0.1%. 0.5 ml light mineral oil was added, and the mixture was sonicated at 60 watts for 10 min. The resulting suspension was centrifuged at 5,000.times.g for 10 minutes to separate phases. After polymerization at room temperature, the mineral oil phase was removed and the polymer was washed with 10 mM Tris-HCl, pH 9.0, containing 4 M urea and 10% SDS for 24 h. The resulting matrix had the form of the interior of an Eppendorf tube.

Detailed Description Text (127):
Acrylamide monomer solution was prepared by dissolving 28.5 g acrylamide and 1.5 g N-N'-methylene bisacrylamide in 100 ml of 4 M urea. 2 mg of the palmitoyl-peptide conjugate molecule of Example 1 was dissolved in 1 ml of the acrylamide monomer solution. Ammonium persulfate and TEMED were added to the solution as catalysts. The final concentration of ammonium persulfate was 0.02%, and the final concentration of TEMED was 0.1%. 0.5 ml light mineral oil was added, and the mixture was sonicated at 60 watts for 4 min. The resulting suspension was centrifuged at 5,000.times.g for 10 minutes to separate phases. After polymerization at room temperature, the mineral oil phase was removed and the polymer was washed with 10 mM Tris-HCl, pH 9.0, containing 4 M urea and 10% SDS for 24 h. The resulting matrix was ground into beads approximately 0.1 mm in diameter.

Other Reference Publication (10):
Sellergren, 1994, "Imprinted dispersion polymers: a new class of easily accessible affinity stationary phases," Journal of Chromatography A 673:133-141.

Other Reference Publication (11):
Spivak and Shea, 1998, "Binding of nucleotide bases by imprinted polymers,"

Macromolecules 31:2160-2165.